CLARIFICATION OF STATEMENTS PROHIBITING THE USE OF HUMAN BODY SUBSTANCES IN THE ALBERTA SCIENCE CURRICULUM

CURRICULUM SUPPORT

Alberta
EDUCATION
1988

Alberta
EDUCATION

Devonian Building, West Tower, 11160 Jasper Avenue, Edmonton, Alberta, Canada T5K 0L2

September 14, 1987

To: Superintendents
School Principals
Science Supervisors and Consultants

RE: USE OF HUMAN TISSUE AND FLUID IN EDUCATION PROGRAMS

Alberta Education has reviewed recent information and concerns expressed by the scientific community about the potential risk of transmitting hepatitis or AIDS through activities that involve the extraction and analysis of samples of human fluid or tissue. The department also has reviewed the current inclusion of such activities as cheek cell scrapes and the taking of blood samples in curriculum. While these activities have been considered acceptable providing that rigorous procedures were followed in the handling, sterilizing and disposal of human tissue and fluid samples and equipment, there is growing concern that even with appropriate procedures a risk to individuals may remain.

Alberta Education after consultation with the Canadian Red Cross Society and the Sexually Transmitted Disease Control Branch of Alberta Community and Occupational Health, believes that the educational value of the activities noted no longer can justify the health risks to those involved. In a time of uncertainty as to the potential risk of infection from hepatitis and AIDS, Alberta Education firmly believes that the safety and well-being of students, teachers and other school staff must be our first consideration. **Therefore, all activities involving the extraction and analysis of samples of human fluid or tissue are now prohibited in Alberta schools.**

Alberta Education will develop guidelines and alternative activities for dealing with components of the educational programs that involve these areas of study. Publishers of learning resources used in schools will be informed of Alberta's position and will be asked to make adjustments accordingly.

For further information or clarification, please contact the Science Consultant in the Alberta Education Regional Office serving your area, or the Curriculum Design Branch.

Sincerely,

Reno A. Bosetti
Deputy Minister

cc: Alberta Teachers' Association
Alberta School Trustees' Association
Alberta Catholic School Trustees' Association

ACKNOWLEDGEMENTS

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GENERAL CLARIFICATION OF STATEMENTS

According to the Alberta Occupational Health and Safety Act and the "in loco parentis" mandate, classroom teachers have the legal obligation to ensure the safety of their classroom for students, other workers and visitors. In laboratory exercises and demonstrations, it is impossible to guarantee an absolutely sterile environment for all students and teachers at all times. It is, then, wise to prohibit the use of potentially dangerous materials in laboratory work and demonstrations done in the classroom. This is particularly important because some infectious organisms have been found to survive long periods of drying.

Current information affirms that the Canadian population has a number of asymptomatic hepatitis B virus carriers as well as individuals infected with the AIDS virus HIV-1, and these numbers are on the increase. It is therefore essential that the educational system inculcate the concept of proper hygiene in the handling of human body substances in accordance with the current understanding of transmission of these infectious agents. Students must be taught that handling body substances of unknown status could endanger their health.

The risk of handling fecal material and urine has at all times been stressed, as a matter of policy, but it is now evident that blood poses an even greater hazard. For this reason it is no longer permissible to expose students to accidental contamination by urine, blood or saliva. Because of the significant risk involved, any experimentation with, or handling of, human tissue is now strictly prohibited.

OVERVIEW

In a survey of the laboratory exercises found in basic texts and laboratory manuals for Alberta senior high biology courses, contentious exercises identified fall into four major groups: those involving blood, urine, saliva and tissue sampling. Appendix I indicates either prohibition or approval with modification of each contentious exercise. General and specific modifications and substitutions are suggested for the four major groups.

Standard laboratory safety practices relating to these areas, and specific procedures for recommended modifications and substitutions, are included in Appendix II and Appendix III respectively. From these guidelines, teachers can generalize the recommendations to other laboratory manuals and texts they choose to use.

SPECIFIC STATEMENTS OF RISK

BLOOD AND URINE

Laboratory Exercises Involving the Chemistry of Urine, and the Analysis of Blood

Because of the risk of contracting hepatitis, AIDS and other sexually transmitted diseases, the use of urine and blood in classroom laboratory exercises is now prohibited. Hepatitis B virus (HBV) and Human Immunodeficiency Virus Type 1 (HIV-1) or Type 2 (HIV-2) are known to be transmitted via contaminated blood contact with open lesions or mucous membranes. Semen, which can be found in urine, is known to have the highest concentration of virus of all human body substances. These facts make the primary materials for these laboratory exercises too risky for their educational value. The risk is increasing at a phenomenal rate as the number of HIV-1 or HIV-2 infected individuals increases in Canada. Even screened negative blood and urine would involve some risk until a more sensitive and reliable test for the virus particles of HIV-1 or HIV-2 is available.

In the future, the bigger school boards may consider setting up central facilities equipped with level 'C' (according to Canadian Medical Research Council guidelines) equipment and air-handling systems for selected groups of students to participate in such activities. Until such facilities are built and equipped, experiments involving human blood and urine must be discontinued in regular classrooms.

Substitution Suggestions

1. Use prepared permanent human blood smear slides for observations of cellular composition.

2. Use of fish, amphibians or reptiles to obtain blood or other tissue samples. The procedure is included in Appendix III.

3. Use videos, films and slides that show typical laboratory procedures involved in human urinalysis, blood typing, blood analysis.

4. Use samples of synthetic urine for laboratory exercises involving testing for the presence of glucose. The procedure is included in Appendix III.

SALIVA

Salivary Amylase Experiments

(Digestion of Starch, Effect of Concentration of Enzyme, Effect of pH, Effect of Temperature on Enzyme Activity.)

Human saliva may contain hepatitis B virus or the tuberculosis bacillus. Saliva is not known to transmit the etiologic agent for AIDS although HIV-1 has been found there. The collection and manipulation of saliva is extremely difficult to execute in a sanitary manner in the classroom setting. It is one matter to have small amounts of saliva soaked into a swab and quite another to collect saliva in test tubes and then manipulate it.

The availability of synthetic enzyme lends itself to safe substitution for salivary amylase in laboratory exercises examining the action of this enzyme. An aqueous solution of ptyalin (amylase) serves well and eliminates the danger of transmission of infection due to careless technique or accident.

Swabs Used in the Mouth

(e.g., The Sensitivity of the Tongue to Different Tastes - *Biology of Ourselves.* Berry, p. 209.)

All used swabs should be placed on disposable absorbent pads and both pads and swabs disposed of, as biohazardous material, by incineration. Surfaces touched by swabs should be decontaminated. Hands should be washed thoroughly both before and after the exercise. Students with open wounds in their oral cavity, or bleeding gums, should not participate in exercises involving swabbing of the mouth area.

Although an obvious safeguard, care must be taken to ensure that <u>fresh</u> swabs are used for each sampling or test taken.

(Surface decontamination and biohazardous waste disposal procedures in Appendix II.)

Testing the Effectiveness of Mouthwash

(e.g., *A Canadian Lab Manual*. Ritter, Drysdale and Coombs, pp. 54-55.)

Students must not be allowed to take each other's samples. The decontamination of desk tops or laboratory bench tops after the exercise is important. The use of absorbent pads is highly recommended for this lab. Disposal of the swabs and these absorbent pads should be by incineration. The students must be instructed to take special precautions to measure the zones of inhibition by the mouthwashes from the bottom of the Petri dish, not by opening the lid. The disposal of the culture dishes should be either by incineration for disposable plastic Petri dishes or by autoclaving for glass Petri dishes. All wastes or contaminated materials must be well labelled with biohazardous materials labels or enclosed in biohazardous logo garbage bags (orange in colour).

The importance of scrubbing the hands at the end of every laboratory session should be stressed.

(Surface decontamination and biohazardous waste disposal procedures in Appendix II.)

PTC Testing

(e.g., The Ability to Taste PTC - *Biology of Ourselves*, Berry, p. 496.)

The chemical phenylthiocarbamide (PTC), although considered a toxic substance, is toxic only at a much higher dosage than one would receive from a simple lick of PTC paper. The LD_{50} ranges from 3 to 40 mg/kg. However, students must be warned and closely supervised to ensure no person tastes a piece of paper previously tasted, and that the papers are disposed of properly. Do not allow students to set used papers down on lab benches or desks, otherwise the surfaces will have to be decontaminated.

Lung Capacity Investigation/Gas Exchange

(e.g., Gas Exchange - *Investigations in Biology*, Benson, Hunt, Lunn, Shostal, p. 76.)

When a spirometer is used a fresh sterile mouthpiece is required for each student. Mouthpieces, if disposable, are placed in biohazard label bag by the student for incineration or autoclaving. If the mouthpiece is to be decontaminated by soaking in 10% bleach solution or Presept solution they should be deposited by the student directly into the solution. When balloons

or straws are used, proper instruction and supervision to avoid sharing of these or saliva becoming airborne is necessary. Proper disposal of balloons or straws and surface decontamination procedures should be followed if they are set down on laboratory surfaces.

(Surface decontamination and biohazardous waste disposal procedures in Appendix II.)

TISSUE

The most common laboratory exercise for observation of typical human cells and their staining properties involves cheek cell scrapings. This practice is no longer acceptable. As a substitute, permanently fixed slides of human tissues can be used to demonstrate human cell characteristics. To illustrate simple staining technique and general animal cell characteristics, a variety of tissues from fish, amphibians or reptiles can be used. Fresh beef kidney from the butcher shop is also a good alternative.

Procedure for preparation of fresh tissue samples from fish, amphibians or reptiles is given in Appendix III along with slide preparation and staining techniques.

Use of Sheep Tissue (Fresh or Frozen)

The use of sheep tissues could be considered biohazardous. There is abundant literature to indicate that the causative organism for Q-fever is widely distributed in sheep flocks in North America and in most parts of the world. The consideration of this known fact is most important for individuals who have heart valve problems, those who are immuno-suppressed, and perhaps those who are pregnant. Consequences of the illness range from fever and symptoms similar to flu, to fatality. Although sheep red blood cells have been found to contain the organism for Q-fever, it has never been reported as causing the infection.

Use of Other Mammalian Tissue (Fresh or Frozen)

It should be ascertained if any student or staff member has a heart valve problem or is immuno-suppressed. If so, these individuals should be excluded from the classroom for any exercise involving handling of fresh or frozen mammalian tissue (infectious agents can become airborne).

If fresh heart-lung plucks, brains or other organs or tissues from the slaughterhouse or from wild mammals are used, extreme care should be exercised to prevent direct contact with the tissues involved. Disposable gloves must be worn, and decontamination of all equipment and surfaces that come into contact with the tissue is essential. Proper disposal of gloves and the tissues as biohazardous material is also required.

(Surface decontamination and biohazardous waste disposal procedures in Appendix II.)

APPENDIX I(a)

PROBLEMATIC LABORATORY EXERCISES FOUND IN BASIC RESOURCES
High School Biology Programs

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* An asterisk indicates those exercises that have been prohibited and for which no modifications have been suggested. All other laboratory exercises noted may be carried out if the appropriate modifications are implemented.

Biology of Ourselves. Gordon S. Berry. John Wiley and Sons Canada Ltd. (1982).

	Activity 2	(p. 38)	Observing Human Cheek Cells
	Activity 5	(p. 209)	The Sensitivity of the Tongue to Different Tastes
	Activity 1	(p. 223)	Coagulation Time
	Activity 2	(p. 223)	Preparing a Blood Smear
*	Activity 3	(p. 224)	ABO Blood Typing Techniques
*	Activity 4	(p. 225)	Rh Blood Typing
*	Activity 5	(p. 225)	The Hemoglobin Content of Blood
	Activity 1	(p. 293)	How Does the Air Entering the Lungs Differ from Air Leaving the Lungs?
	Activity 3	(p. 294)	Measuring the Capacity of the Lungs
	Activity 1	(p. 322)	The Salivary Digestion of Starch (Part I)
*	Activity 1	(pp. 392-393)	Analysis of Urine
	Activity 1	(p. 496)	The Ability to Taste PTC

Laboratory Biology: Investigating Living Systems. Kaskel, Hummer, Kennedy, Oram. Charles E. Merrill Publishing Co. (1983) Canadian SI Edition.

	Lab 9	(pp. 35-38)	Proof of Enzyme Action
	Lab 12	(p. 48)	The Basic Unit of Life Part B: Cell Membrane and Cytoplasm
*	Lab 27	(pp. 105-108)	A Human Variation with Possible Adaptive Value
*	Lab 64	(p. 250)	Blood, Part D: Plasma
	Lab 65	(pp. 251-254)	Lung Capacity
*	Lab 66	(pp. 255-258)	Urinalysis

Biology - A Canadian Laboratory Manual. R. Ritter, B. Drysdale, D. Coombs. GLC Silver Burdett.

	Lab 4	(p. 48)	Microscopic Investigation of Animal Tissue
	Lab 6	(pp. 54-55)	Testing the Effectiveness of Mouthwash
*	Lab 52	(pp. 223-226)	Microscopic Investigation of Urine

APPENDIX I(a) (cont'd)

Biology - A Canadian Laboratory Manual. Teacher's Manual (shrink-wrapped, 3 hole punched). R. Ritter, B. Drysdale, D. Coombs. GLC Silver Burdett.

*		(pp. 151-160)	Blood Typing
		(pp. 161-169)	Identification of Blood Cells
*		(pp. 170-175)	The Genetics of Human Blood

Investigations in Biology. (First edition, 1977) Benson, Hunt, Lunn, Shostal, Addison-Wesley.

	Investigation 16	(p. 35)	The Influence of pH on Enzyme Activity
	Investigation 17	(p. 37)	The Effect of Temperature on Enzyme Activity
	Investigation 27	(p. 60)	Dissection of a Mammalian Heart
*	Investigation 29	(p. 66)	Blood Antigens
	Investigation 31	(p. 70)	Identification of Human Blood Cell Types
*	Investigation 32	(p. 73)	Immune Reactions
	Investigation 33	(p. 76)	Gas Exchange (spirometer) Sterile Disposable Mouthpieces or Disinfect
	Investigation 34	(p. 79)	Exhalation of Carbon Dioxide (straws - disposable)
*	Investigation 38	(p. 87)	Composition of Urine
	Investigation 46	(p. 110)	The Eye

Investigations in Biology. (Second Edition, 1986) Lunn, Hunt, Shostal, Addison-Wesley.

	Investigation 37	(p. 98)	Dissection of a Mammalian Heart
*	Investigation 39	(p. 104)	Blood Antigens
	Investigation 41	(p. 108)	Identification of Human Blood Cell Types
	Investigation 43	(p. 114)	Gas Exchange
	Investigation 44	(p. 117)	Exhalation of Carbon Dioxide
	Investigation 47	(p. 124)	Kidney Structure
*	Investigation 48	(p. 125)	Composition of Urine
	Investigation 49	(p. 128)	Nervous Coordination: Structure
	Investigation 53	(p. 139)	Taste Receptors
	Investigation 54	(p. 142)	The Eye

APPENDIX I(b)

PROBLEMATIC LABORATORY EXERCISES FOUND IN BASIC RESOURCES
Junior High Science Programs

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* An asterisk indicates those exercises that have been prohibited and for which no modifications have been suggested. All other laboratory exercises noted may be carried out if the appropriate modifications are implemented.

Challenges to Science - Life Science. William Smallwood, McGraw-Hill Book Company (1973).

	Lab. Activity	(p. 64)	What are Organisms Made Of? Procedures 3, 4, 5, 6
*	Lab. Activity	(p. 172)	What ABO Blood Type Genes Could You Have?

Life Science, A Problem-Solving Approach. Carter, Goodman, Hunter, Schelske, Ginn and Company (1971).

	Problem 6-2	(p. 97)	What are the parts of a human cheek cell?
	Problem 9-3	(p. 163)	Where can microorganisms be found?
	Problem 10-1	(p. 173)	What are some types of bacteria colonies?
	Problem 10-2	(p. 177)	What effects do antiseptics have on bacterial growth?
	Problem 10-3	(p. 181)	What effect does ultra violet light have upon bacterium *Serratia marcescens*?

APPENDIX II

STANDARD LABORATORY SAFETY PRACTICES

SURFACE DECONTAMINATION PROCEDURES

Standard Procedure

Wipe area down well with freshly made 10% bleach solution (approximately 10,000 ppm or 10,000 mg/L of available chlorine).

Special Procedure (areas where tuberculosis occurs)

Wipe area down well with Presept, a Johnson & Johnson product. This product will destroy tuberculosis bacilli as well as the hepatitis and AIDS viruses.

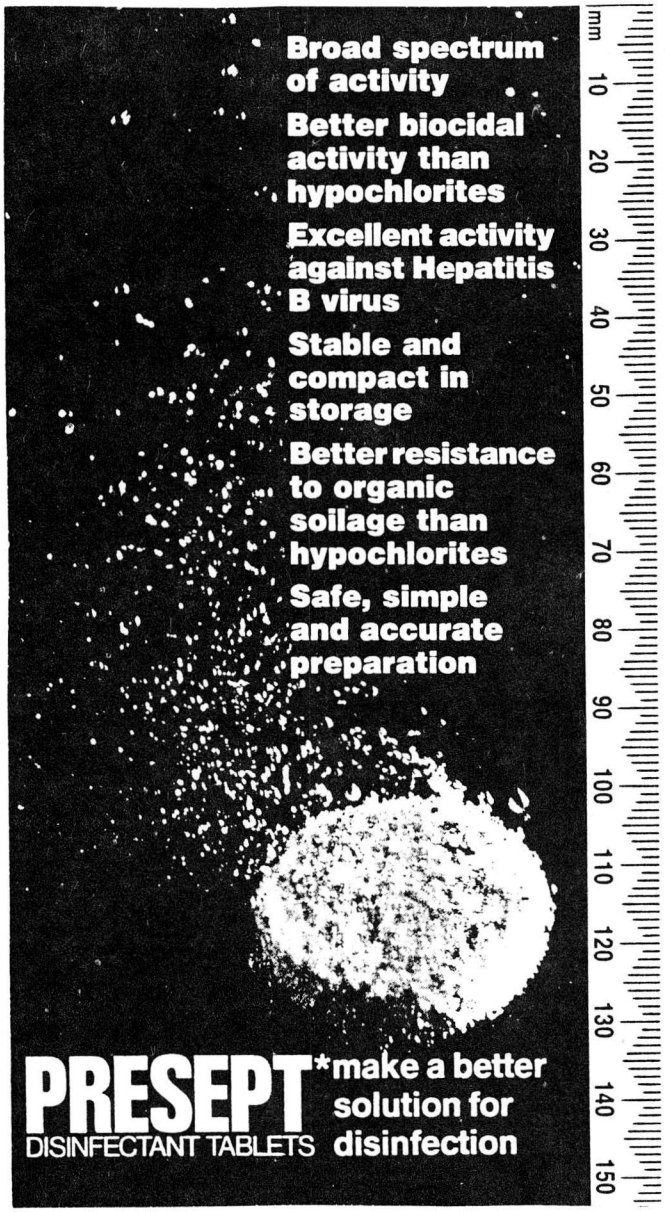
Presept (Johnson & Johnson) available from:

Canada Medical Ltd.
Edmonton, Alberta
Toll free number: 1-800-232-7286
Local number: 465-2020

Prairie Medical Ltd.
Edmonton, Alberta
Toll free number: 1-800-661-7347
Local number: 451-2910

Prairie Standard Medical
Calgary, Alberta
Toll free number: 1-800-661-8423
Local number: 279-3550

Presept product information is on the following page.



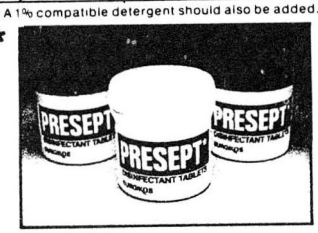
Dilution instructions for PRESEPT* Disinfectant Tablets

Disinfection of	Required Concentration	Dilution Rates			Additional Instructions
		0.5g tablets	2.5g tablets	5.0g tablets	
Blood spillage †	10,000 ppm	18 tablets in 0.5 l water	7 tablets in 1 l water	9 tablets in 2.5 l water	Pour over blood. Using gloves, wipe up with disinfectant-saturated disposable cloth.
Pipette jars †	2,500 ppm	9 tablets in 1 l water	9 tablets in 5 l water	9 tablets in 10 l water	Drop tablets into water-filled pipette jar. Discard daily ‡.
General laboratory/ environmental use †	1,000 ppm	4 tablets in 1 l water	4 tablets in 5 l water	3.5 tablets in 10 l water	Wipe down surfaces with disposable disinfectant-saturated cloth.
Stainless steel instruments	560 ppm	1 tablet in 0.5 l water	1 tablet in 2.5 l water	1 tablet in 5 l water	Immerse for 1 hr.
Baby bottles/nipples stainless steel utensils, porcelain, glassware, rubber and plastic tubing	140 ppm	1 tablet in 2 l water	1 tablet in 10 l water	1 tablet in 20 l water	Immerse for 1 hr.
Eating utensils and crockery	140 ppm	1 tablet in 2 l water	1 tablet in 10 l water	1 tablet in 20 l water	Rinse for 1-2 mins.
Soiled linen	140 ppm	1 tablet in 2 l water	1 tablet in 10 l water	1 tablet in 20 l water	Immerse for 1 hr. prior to washing
Infected linen	140 ppm	1 tablet in 2 l water	1 tablet in 10 l water	1 tablet in 20 l water	As above
Work surfaces, cupboards, floors, etc.	140 ppm	1 tablet in 2 l water	1 tablet in 10 l water	1 tablet in 20 l water	Wash down
Dishcloths, mops, etc.	60 ppm	1 tablet in 4.6 l water	1 tablet in 23 l water	1 tablet in 46 l water	Soak to bleach clean and deodorize

†Based on studies available upon request. ‡ A 1% compatible detergent should also be added.

PRESEPT*
DISINFECTANT TABLETS
make a better solution for disinfection

The infection control company
SURGIKOS
a Johnson & Johnson company



Reorder Codes:
PB05, 0.5g active ingredient, 6x600 tabs.
PR25, 2.5g active ingredient, 6x100 tabs.
PW50, 5.0g active ingredient, 6x50 tabs.

Surgikos Canada Inc.
1355 Lansdowne St. W.
Peterborough, Ont. K9J 7X2

Courtesy of Surgikos, a Johnson & Johnson Company.

Labelling and Proper Disposal Technique

All biohazardous garbage to be incinerated or autoclaved must be clearly identified by placing in special orange biohazard logo garbage bags or by attaching a clearly visible biohazard label. For materials to be autoclaved, either temperature sensitive autoclave tape or bags with temperature sensitive biohazardous logos should be used. The autoclave tape or logo changes colour when autoclaved, clearly indicating the contents have been sterilized. Sharp instruments such as lancets, syringes, scalpel blades or razor blades must be placed in appropriate biohazard canisters. When full, these canisters should be incinerated or autoclaved and then disposed of in a landfill site.

Label Sample:

Biohazardous labels available from:

Fisher Scientific
Edmonton, Alberta
Local number: 483-2123
Toll free number: 1-800-661-9981

Lab Safety Supply
P.O. Box 1368
Janesville, WI. 53547-1368
U.S. Toll free number: 1-800-356-0783

Levitt Safety
Calgary, Alberta
Local number: 243-4273

Safety Supply Canada
Toll free number: 1-800-387-4558

Autoclave tape (sterilizer indicator tape) available from:

Fisher Scientific
Edmonton, Alberta
Local number: 483-2123
Toll free number: 1-800-661-9981

NOTE: Also available from other biological companies.

Garbage bags with Biohazardous Logo - bright orange, available in a variety of sizes, autoclave indicating or plain logo.

Fisher Scientific
Edmonton, Alberta
Local number: 483-2123
Toll free number: 1-800-661-9981

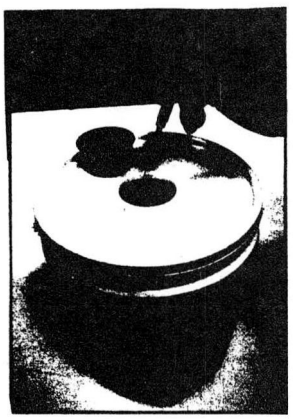
Chemonics
Edmonton, Alberta
Local number: 451-0665
Calgary, Alberta
Local number: 250-1142 (No toll free number)

Sargent-Welch
Calgary, Alberta
Local number: 291-3090
Toll free number: 1-800-482-6112

Biohazard Canisters for Sharp Instrument Disposal available from:

Canada Medical Ltd.
Edmonton, Alberta
Local number: 465-2020
Toll free number: 1-800-232-7286

PRO-WESTERN
SHARPS DISPOSAL SYSTEM



MEETS C.S.A.
SPECIFICATIONS

MANUFACTURED BY:
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30 RIEL DRIVE, P.O. BOX 261 ST. ALBERT, ALBERTA T8N 1N3
TEL. (403) 459-4491 TELEX 037-41600

SAFETY

- Meets CSA Specification Z317.10 - M1981
- Color Coded Containers Printed With BioHazard Symbol
- Puncture Resistant Heavy Wall Containers
- Tight Sealing Lids Preventing Contents From Spilling
- Non-Porous Plastic - DOES NOT CONTAIN PVC
- Can Be Incinerated - No Toxic Fumes

CONVENIENT

- Multiple Sizes Which Can Be Used Throughout Hospitals
- Easy To Set Up and Discard
- Easy To Identify Color Coded Printed Containers

ECONOMICAL

- Canadian Manufactured Product
- All Product Standard Stock For Immediate Release
- Nestable Containers Reduces Storage Space

Courtesy of Pro-Western Plastics Ltd.

Culture Plate Procedures

Use of agar plates is a simple method of culturing bacteria. When classroom procedures involve random sampling of unknown materials, caution must be taken to avoid direct contact with cultured colonies in order to avoid accidental exposure to pathogenic organisms.

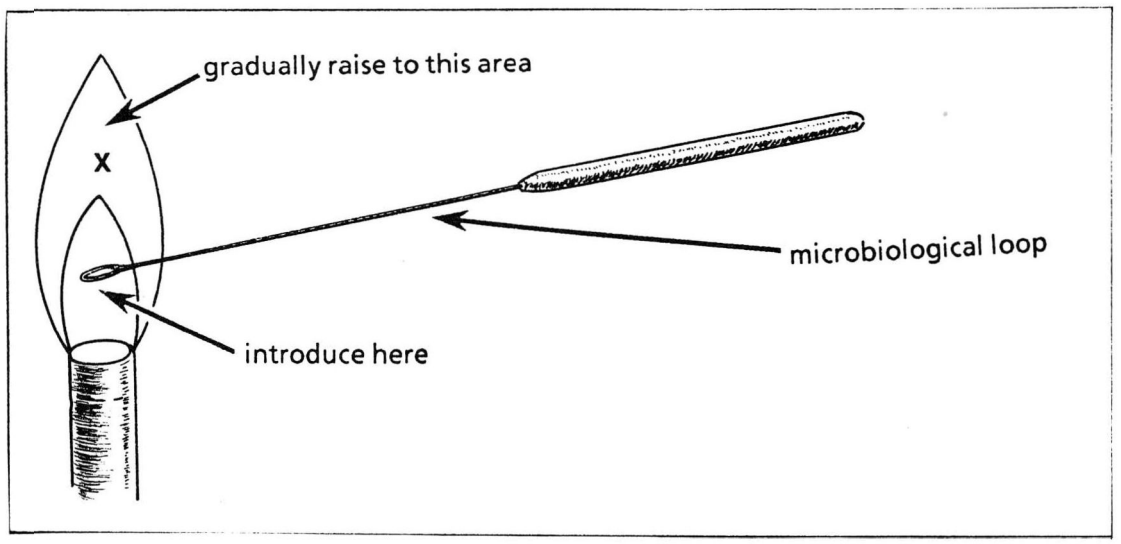
Immediately after streaking, swabs used for sampling should be disposed of in a biohazard garbage bag, for incineration or autoclave sterilization. Bench tops should be surface decontaminated at the end of each laboratory session. (Surface decontamination procedures in Appendix III.) Staff and students should develop the habit of washing their hands thoroughly with soap and water at the end of the exercise.

As soon as the agar surfaces of the culture plate have been streaked, the lid should be taped to the bottom half of the Petri plate to prevent accidental contact with cultured organisms.

Clearly labelled Petri plates should be incubated upside down, in stacks which are then secured in a plastic bag.

All observations of cultured colonies are to be done <u>without</u> opening the taped plates. Colonies are clearly visible through the lid, and zones of inhibition can be measured from the bottom.

If samples of cultured colonies are to be taken for fixing, staining and microscopic observation, the proper sampling techniques using a microbiological loop must be followed. Lid lifting is minimized, with loop being flamed before sample-taking and after placement of sample onto the slide. Proper flaming of the loop involves introducing the loop into the cooler, middle cone of the Bunsen burner and then slowly raising it into the hotter, more exterior region of the flame. (See diagram below.) This prevents the spattering of possible pathogens before they are killed by the heat.



APPENDIX III

a) Canadian Council for Animal Welfare - General Rules and Regulations.

b) Procedures for Obtaining Fresh Tissue Samples from Fish, Amphibians and Reptiles.

c) Procedure for Making Synthetic Urine (for glucose test only).
Refer to Teacher's Guide: *Biology - A Canadian Laboratory Manual*. Ritter, R.; Drysdale, B.; D. Coombs. GLC Silver Burdett. (Laboratory 53, page 137: Insulin and Blood-Sugar Regulation.)

APPENDIX III(a)

CANADIAN COUNCIL FOR ANIMAL WELFARE - GENERAL RULES AND REGULATIONS

The following excerpts from:

Canadian Council on Animal Care
Guide to the Care and Use of Experimental Animals. 2 vols.
Ottawa, Ont.: CCAC, 1980-1984.

Copies may be obtained from the:

Canadian Council on Animal Care
1000 - 151 Slater Street
Ottawa, Ontario
K1P 5H3
Telephone: (613) 238-4031

Approximate cost of the two volumes is $9.00 including postage.

Preface

The Canadian Council on Animal Care (CCAC) is an independent, incorporated body, supported financially by the Natural Sciences and Engineering Research Council (NSERC) and the Medical Research Council (MRC).

An Executive Director and a Director of Assessments head the CCAC secretariat which, with the assistance of its committees, serves as a resource and information centre for those using experimental animals, as well as for the general public. Assistance is also offered to persons responsible for the care and use of animals in pre-university educational programs.

The fundamental concept on which animal care in Canada and its surveillance by CCAC is based, is that of control from within an institution, exercised by the scientists themselves in association with non-animal users from other faculty departments, and representatives of the community. This implies the creation of local animal care committees with specific authority, terms of reference and responsibilities, for assuring that all matters concerning the use and day-to-day care of experimental animals within institutions are carried out in accordance with the recommendations of CCAC.

A 110-member pool of scientists from universities, government, and the pharmaceutical industry, aided by representatives appointed by the Canadian Federation of Humane Societies (CFHS), is responsible for the implementation of the CCAC nation-wide assessment program. This system of periodic surveillance provides support to institutional animal care committees, uniformity of national standards and a means of keeping the agencies represented on the CCAC properly informed. Assessment panel and committee members are chosen for their expertise in a variety of fields, and their leadership and concern in matters of animal care and use. They serve without remuneration.

Both the French and English editions of this guide have become internationally known. These have provided a basis for assessment evaluations across Canada. Moreover, adherence to the principles enuciated in the guide, and a satisfactory assessment, are now required by all major national granting agencies. This is also a requisite for the awarding of contracts involving research animals by the Federal Departments of Health and Welfare, Agriculture, Environment and National Defence.

Legislation

CCAC's system of self-regulation by and with the full support of the scientific community, government agencies and the Canadian Federation of Humane Societies, is designed as an alternative to the necessity for national legislation of the care of experimental animals.

Federal legislation controlling the conditions under which animals must be transported is included in the "Animal Disease and Protection Act".

The procurement of animals (dogs and cats) for research teaching and testing is regulated in Alberta, Ontario and Saskatchewan. In addition, the Alberta and Ontario acts regulate the conditions under which animals are maintained and used for these purposes, through local animal care committees.

The Scientist-Teacher

The scientist-teacher should have knowledge of the characteristics, care and handling of the species he or she proposes to use and be committed to comply with the guidelines for care and ethical use of animals as contained in this guide.

The ultimate responsibility for the prevention of pain and discomfort in the experimental situation lies with the investigator.

Pre-University

There is sometimes a need for animals in the classroom at the pre-university level. This makes it possible to teach principles of animal care to students, so that they may better understand the normal functional and behavioural aspects of animals, as well as to inculcate a proper sense of responsibility toward animals and their care.

Unlike the investigator or teacher at the university level, students and many teachers at the pre-university level do not have the necessary training or experience to conduct animal-based research. Therefore, they should not engage in studies which would subject an animal to pain or discomfort, or interfere with its health and well-being. Comments on experimental studies involving animals at the pre-university level, as well as the names of qualified experts on animal care and use, may be obtained by writing to CCAC.

This guide has been developed primarily for use at the post-secondary level. However, its principles apply equally well to pre-university work with experimental animals. Those planning to undertake such work should therefore become familiar with the contents of this guide and adhere to the guiding principles of the CCAC and the Youth Science Foundation (YSF).

The Youth Science Foundation (YSF), Suite 904, 151 Slater Street, Ottawa, Ontario K1P 5H3, is the umbrella organization responsible for all out-of-school science activities in Canada. Its objective is to develop scientific awareness among pre-university students as well as to encourage scientific literacy with the Science Fair Program, one of its principal activities.

YSF programs are directed primarily toward the English-speaking students in Canada's ten provinces and territories. The Conseil de développement du loisir scientifique (CDLS), 4545, rue Pierre du Coubertin, C.P. 1000, Succursale M, Montréal, Québec, H1V 3R2, by mutual agreement with the YSF, is responsible for similar activities for Francophone students, primarily in the province of Quebec.

When the Canadian Council on Animal Care was established in 1968, the council, together with representatives from the Canadian Veterinary Medical Association in concert with the Youth Science Foundation, recognized the importance of well-conceived science fair projects involving live animals. Over the years, attempts were made to ensure development of proper scientific investigational attitudes as well as respect for living things. As these early efforts proved largely unsuccessful, increasingly stringent requirements were made. Finally, in May 1975, the regulations now in effect, developed by the Animal Care Committee of the Youth Science Foundation and CCAC, were adopted.

The Canadian Council on Animal Care, following the acceptance of the regulations for animal experimentation in science fairs and their implementation, prepared guidelines governing the use of animals in the classroom at the pre-university level for use by departments of education and boards of education across Canada. Their aim was to ensure the existence of adequate safeguards for the proper care and use of animals in experimentation in the classroom.

Suggestions for experiments involving animals at the pre-university level are available from CCAC.

Additionally, collaboration has been undertaken with various high school educational authorities in the preparation of manuals and course outlines involving animals and teaching programs at the pre-university level.

Guiding Principles Governing the Use of Animals in the Classroom at the Pre-University Level

I. Purpose

These guiding principles have been prepared by the Canadian Council on Animal Care. They are recommended for use by departments of education and boards of education across Canada in order to ensure adequate safeguards exist for the proper care and use of animals in experimentation in the classroom, by the schools in their jurisdiction.

These guidelines are not for use by students preparing projects for exhibit in science fairs. Students preparing projects for science fairs must adhere to the Youth Science Fair Regulations for Animal Experimentation, as prepared and distributed by the Youth Science Foundation, Suite 302, 151 Slater St., Ottawa, Ontario K1P 5H3.

II. Philosophical Considerations

Biological experimentation involving animals in the classroom is essential for an understanding of living processes. Such studies should lead to a respect for all living things. All aspects of the study must be within the comprehension and capabilities of the students undertaking the study.

Lower orders of life are preferable subjects for experimentation at the pre-university level. Such lower orders as bacteria, fungi, protozoa, and insects can reveal much basic biological information; they should be used for experimentation, wherever and whenever possible.

III. Care of Experimental Animals

The care of experimental animals in the school should embody the principles laid down in this guide.

The following principles are necessary in order to provide optimal animal care:

a) The maintenance of animals in a classroom shared by students is not recommended on a long-term basis. Therefore, animal quarters specifically for housing of animals should be provided.

b) All experimental animals used in teaching programs must be properly cared for. Animal quarters should be made comfortable by provisions for sanitation, protection from the elements and sufficient space for normal behavioural and postural requirements of a

species. The living quarters shall have surfaces that may be easily cleaned; good ventilation and lighting, well regulated temperatures; and cages of sufficient size to prevent overcrowding. Animals must be protected from direct sunlight or other environmental factors which may disturb their well-being.

c) Food should be palatable, and of sufficient quantity and balance to maintain a good standard of nutrition. Foods intended for animals shall not be allowed to fall below the recommended maintenance level of nutrition. Clean drinking water shall be available at all times. Containers for food and water should be of a design made specifically for that purpose.

d) Colonies and animal quarters shall be supervised by a science teacher experienced in animal care. The students and other animal care staff shall be trained and required to handle the animals gently and humanely.

e) All animals must be disposed of humanely. If any animal has to be destroyed, an approved humane method must be carried out by an adult experienced in the use of such procedures.

f) The use of animals must comply with existing local, provincial or federal legislation.

g) The procurement and use of wild animals and birds must comply with the Migratory Birds Convention Act of Canada, the Convention on International Trade on Endangered Species of Wild Fauna and Flora (ratified by Order in Council July 3/75) as well as any existing legislation at the provincial level concerned with wild animals and exotic species.

IV. Experimental Studies

1. All experiments should be carried out under the supervision of a competent science teacher. It is the responsibility of the qualified science teacher to ensure that students have the necessary comprehension for the study to be undertaken.

2. Students should not be allowed to take animals home for the purpose of carrying out experimental studies. All studies involving animals must take place in a suitable area in the school.

3. All students carrying out projects involving vertebrate animals must adhere to the following guidelines:

   A. No experimental procedures shall be attempted on a vertebrate animal that could subject it to pain or distinct discomfort, or interferes with its health.

   B. Students shall not perform surgery on vertebrate animals.

   C. Experimental procedures shall not involve the use of:
      a. microorganisms that can cause diseases in man or animals.
      b. ionizing radiation.
      c. cancer producing agents.
      d. drugs or chemicals at toxic levels.
      e. alcohol in any form.
      f. drugs that may produce pain.
      g. drugs known to produce adverse reactions, side effects, or capable of producing birth deformities.

   D. Experimental treatments should not include electric shock, exercise until exhaustion, or other distressing stimuli.

   E. Behavioural studies should use only reward (positive reinforcement) and not punishment in training programs.

   F. If egg embryos are subjected to experimental manipulations, the embryo must be destroyed humanely two days prior to hatching. If normal egg embryos are to be hatched, satisfactory humane considerations must be made for disposal of the young birds.

4. The use of anaesthetic agents by students is not recommended and in the case of some anaesthetics not permitted by law.

5. Information on the care, housing and management of individual species, as well as suitable experiments for use at the pre-university level, may be obtained from the Canadian Council on Animal Care, 1000-151 Slater St., Suite 1105, Ottawa, Ontario. K1P 5H3.

Youth Science Foundation Regulations for Animal Experimentation in Science Fairs

1. Biological experimentation is essential for an understanding of life processes. Such studies should lead to a respect for all living things. Capable students, anxious to pursue a career in biological sciences, must receive the necessary encouragement and direction. **All aspects of the project must be within the comprehensions and capabilities of the student undertaking the study.**

2. Lower orders, such as bacteria, fungi, protozoa and insects can reveal much basic biological information. If experiments are to be conducted on living subjects for science fair projects then only lower orders of life may be used.

3. **Vertebrate animals are not to be used in experiments for projects or for science fairs,** with the following exceptions:

   A. Observations of normal living patterns of wild animals in their free habitats or in zoological parks, gardens or aquaria.

   B. Observations of normal living patterns of pets, fish or domestic animals.

4. **No living vertebrate animal shall be displayed as exhibits in science fairs.**

5. Cells such as red blood cells, other tissue cells, plasma or serum purchased or acquired from biological supply houses or research facilities, may be used in science fair projects.

6. Observational type studies on only chicken egg embryos may be used in science fair projects. If normal egg embryos are to be hatched, satisfactory humane considerations must be made for disposal of the chicks. If such arrangements cannot be made then the chicken embryos must be destroyed on the 19th day of incubation. No eggs capable of hatching may be exhibited in science fairs.

7. All experiments shall be carried out under the supervision of a competent science teacher. It shall be the responsibility of the qualified science teacher to ensure the student has the necessary comprehension for the study to be undertaken. Whenever possible specifically qualified experts in the field shall be consulted.

Euthanasia

The term "euthanasia" is used to describe the process whereby an animal is killed using recognized acceptable humane techniques. By derivation, it means "easy death" and thus carries the explicit implication of a quiet, painless death without fear or anxiety.

The most important criterion of acceptance of a euthanasia method as a humane process is that it have an initial depressive action on the central nervous system (CNS) to ensure immediate insensitivity to pain.

It is important to recognize that some methods of euthanasia, which cannot be made aesthetically pleasant, such as decapitation or stunning with exsanguination, may nonetheless be humane in terms of the above criterion. This concept is important to keep in mind when deciding on the method of euthanasia to be used. The choice must be based on the sensibilities of the animal to be killed rather than the sensitivities of the observer or operator, although the latter should not be disregarded.

APPENDIX III(b)

PROCEDURES FOR OBTAINING FRESH TISSUE SAMPLES FROM FISH, AMPHIBIANS AND REPTILES

> When anesthesia or euthanasia is required to obtain tissue samples these procedures are to be performed by the teacher before the laboratory activity commences. Students should not perform or observe these procedures.

Handling, anesthesia and euthanasia methods specific to fish, amphibians and reptiles as prescribed by the CCAC, are as follows:

FISH

Handling Methods

1. Physical Restraint

Fish are covered with a thin, delicate cuticle, and damage to the skin represents a break in the osmotic barrier between fish and water. When handling fish, therefore, great care must be taken to avoid damaging the skin. The hands should always be wet, as should nets and all other materials coming into contact with the fish. In order to avoid abrasions to the skin of struggling fish, netting materials should not be too coarse or hard.

2. Chemical Restraint

Anesthesia, administered via the water, may be desirable if a lot of handling is necessary. Numerous anesthetics are available, including carbon dioxide and benzocaine; however, the one in most common use is tricaine methane sulphonate (MS-222) at a concentration of 1/12,000-1/25,000, depending on the depth of anesthesia and speed of induction required.

3. Bleeding

Blood samples may be obtained from anesthetized fish with no detrimental long-term effects. The two main sites for this are the caudal vein and the heart, with the former being preferable. The dorsal aorta, running along the roof of the mouth, is an alternative sampling site in salmonoids.

4. Euthanasia

Euthanasia may be accomplished by an overdose of anesthetic or by spinal severance just behind the head. For large fish, or where anesthetics are to be specifically avoided, a sharp blow on the top of the head, while firmly holding the fish out of water, is also considered a humane method.

AMPHIBIANS AND REPTILES

Frogs and toads should be picked up by the hind legs or, with experience, by the body; use of a cloth may be helpful. When it is necessary to pick up an axolotyl, or other urodeles with external gills, particular care must be taken not to put pressure on them.

Because of their permeable skin, amphibians can be easily anesthetized by placing them in a solution of anesthetic. Tricane methane sulfonate (MS-222), as a 0.01% aqueous solution, will produce anesthesia within a few minutes, depending on the size of the amphibian.

Hypothermia is often recommended as a form of anesthesia in cold-blooded animals. Placing the animal in a refrigerator or ice water bath immobilizes it. A state of unconsciousness appears to be produced; however, it is not known whether this is actually so and whether analgesia is really induced. For these reasons, hypothermia should be used only for restraint for relatively non-painful procedures and not as an anesthetic for major surgery.

Blood Smears

A goldfish, frog, salamander, newt, lizard, turtle or tortoise could serve as a source of fresh blood for blood smear preparation. A fixed permanent human blood smear can be used for comparison.

The same procedure previously prescribed in basic resources for human blood smear preparation, fixing and staining should be followed, substituting the selected animal's blood. Wright's stain is the most commonly used and would work well. Other stains that might be tried are Leishman-Giemsa, Lugol's, Jenner-Geimsa, or Kleinberg-Nobel.

To obtain blood from a fish, amphibian or reptile, anethesia by immersion in the appropriate solution concentration of MS-222 is necessary. The solution is prepared by dissolving the appropriate

amount of MS-222 (TMS) crystals in water. When preparing the solution, teachers or laboratory assistants should wear disposable gloves, and a mask over the nose and mouth area. When immersing or removing the specimen from the anesthetic bath gloves must be worn. Fresh solution is required.

MS-222 (tricaine methane sulphonate or TMS) available from:

Syndel Laboratories Ltd.
Vancouver, B.C.
Toll free number: 1-800-663-2282

Safe disposal of used MS-222 solution requires that vermiculite be used to absorb the liquid, that it be bagged and labelled biohazardous, and then incinerated.

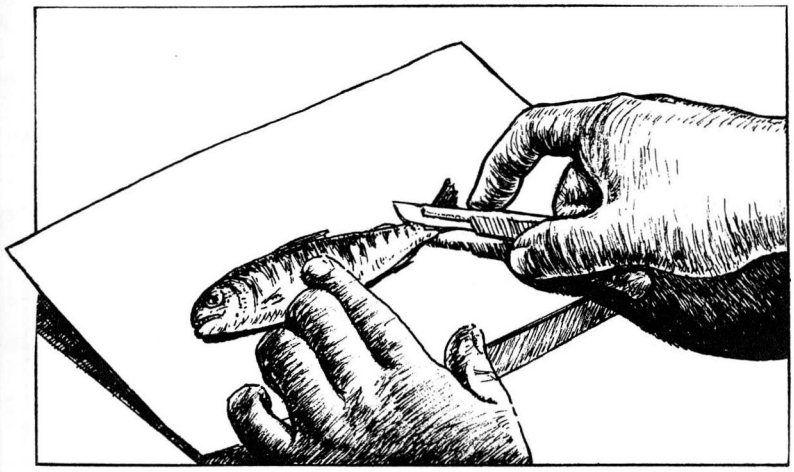
Anesthetizing amphibians and reptiles for blood sampling requires a 1:2000 solution of MS-222. Once anesthethized by immersion, blood may be obtained from frogs or toads by opening the mouth and extending the tongue forward, revealing the floor of the oral cavity. Clean out the mucus and use a lancet to pierce the lingual vein posterior to the hyoid cartilage. Blood can then be drawn up in capillary tubes containing a small amount of 10% citrate solution to prevent clotting. Blood can be distributed to the students' slides from the capillary tube. Bleeding will stop spontaneously when the tongue is placed back in the mouth. Put the animal back in fresh water immediately to recover.

Newts' or salamanders' blood can be drawn by piercing a toe with a lancet and collecting the flowing blood in 10% citrate solution capillary tubes as described above. The best spot for piercing a reptile's toe is at the junction of the terminal phalanx and the claw. Return the animal immediately to fresh water to recover. Bleeding of the animal will stop spontaneously.

A medium-sized goldfish can be immersed in a 1.0% solution of MS-222 which will kill it painlessly. A single goldfish can then serve to supply blood for the class as well as be a source of other tissue samples for microscopic examination. Blood may be obtained by removing the tail with a sharp knife and collecting blood from the dorsal aorta with capillary tubes containing small amounts of 10% of citrate solution (see diagram following). Various other tissues can also be collected for examination. Epithelial cell scrapings from inside the cheek of the fish can replace human cheek cell scrapings. Cell squash preparations for gill tissue, liver, pancreas and other soft

tissue areas can serve to show cellular differentiation. Thin cross-sections of muscle tissue and gut cross-sections can also be attempted. Methylene blue stain is effective for these temporary preparations. Gross gill structure, scale overlap and structure and tail capillary structure can also be observed.

Lancets must be properly disposed of in biohazard canisters. All used slides and surfaces must be decontaminated. Procedures for both are contained in Appendix III.



Make a swift cut completely through the fish at the position shown, slightly forward of the narrowest section between the trunk and tail.

To obtain blood easily from the caudal cut, tip the fish as shown and gently squeeze the anterior portion while touching the capillary tube to the dorsal aorta.

REFERENCES

Anderson, Douglas P. **Fish Immunology**. T.F.H. Publications, Inc. Ltd. (1974).

Canadian Council on Animal Care. **Guide to the Care and Use of Experimental Animals**. 2 Vols. Ottawa, Ont.: CCAC, 1980-1984.